

Abnormal Gene Expression of Four Genes in Cells from Family Members of Hereditary-type Retinoblastoma Patients relative to Normal Individuals

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Abstract

Recently, we have demonstrated an abnormal gene expression profile in unaffected parents in five families of patients with hereditary-type Retinoblastoma (RB). In other studies we also found that cells from these same parents were clearly hypersensitive with respect to the levels of γ -H2AX foci in a low dose-rate irradiation assay in non-cycling G₀ cells and in a G₂/M assay, both of which reflect defects in the processing of DNA double strand breaks. The possibility of an enhanced germline mutation rate in these parents was suggested, perhaps resulting from some mild defect in genome maintenance. In this study, we further examined the differences in expression of a few of the genes identified in the earlier studies in cells from members of these five RB families relative to cells from normal individuals. Polo-like kinase 1 (*PLK1*) is a critical regulator of the cell cycle which governs multiple events associated with G₂-M-phase progression and exhibits aberrant expression in various human tumors. By use of quantitative reverse transcription-PCR (RT-PCR) measurements, significant differences in the expression of *PLK1* were observed for cells from members of these RB families compared with the clinically apparently normal human fibroblast controls. Three independent experiments were carried out for each non-cycling confluent cell strain kept in a G₀ state. Expression levels were normalized to the housekeeping gene, *Actin*, and the *PLK1* expression was from 2- to 5-fold greater than that for the samples from normal control individuals. Further study of the other three genes identified in the expression array studies including insulin-like growth factor-binding proteins: IGFBP-5, IGFBP7 and type IV collagen α 1 (COL4A1) are also being investigated, and the preliminary RT-PCR data for samples from the RB families for these 3 genes also appear to display a distinctive expression pattern relative to samples from normal control individuals.

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